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THE DEVELOPMENT OF *FASCIOLOPSIS BUSKI* LANKESTER

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Fasciolopsis buski is an important human parasite which has been reported from China, India, Assam, Bornea, Sumatra, Cochin China and Tonkin. It is most frequently found in China where it has been reported from Canton, Hongkong, Swatow, Shanghai and other places. According to Goddard (1920) there is a very important endemic center of this parasite in the region of Shaohing in the Chekiang province. In Formosa it is not infrequently found in pigs, but no human cases have as yet been reported.

I obtained specimens of *Fasciolopsis buski* from native pigs in Formosa which measured 25 to 40 mm. in length and 10 to 17 mm. in width. Eggs from these individuals were incubated and free swimming miracidia obtained. Penetration of these miracidia was obtained into *Planorbis coenosus* and the development of the cercaria was studied. Later encysted cercariae were fed to experimental animals and the stages in development to the adult condition were traced. These studies were carried on over a period of five years, and an outline of them has already been published (Nakagawa, 1921). In the present paper I will give a more detailed account of the stages in development.

The development of the eggs of *Fasciolopsis buski* resembles that described by Leuckart for the eggs of *Fasciola hepatica*. The eggs (Fig. 1) fresh from the uteri of the worms are a light yellowish brown in color and measure 0.12 to 0.14 mm. in length (average 0.13 mm.) and 0.07 to 0.09 mm. in width (average 0.08 mm.). A small operculum is present. When these eggs are placed in a small glass dish and the water changed each day, the fertilized egg cell undergoes cleavage and the miracidium soon begins to take form. At first the development is rather slow but later becomes more rapid. During the development of the miracidium the surrounding yolk cells begin to dissolve and gradually disintegrate into what appear to be drops of oil. The end of the developing embryo toward the operculum is broader and develops into the anterior end of the miracidium, while the opposite end early becomes somewhat pointed. As the length of the embryo increases, the surface of its body becomes ciliated and a small protuberance appears at its anterior tip. Just back of this there develops an X-shaped pigmented eyespot. About this time slight extensions and contractions of the body of the embryo can be noted. These movements gradually increase until the miracidium finally pushes off the operculum and escapes into the water. Figure 2 shows the fully developed miracidium in the egg. The structure of the miracidium (Fig. 3) is very similar to that of the miracidium of *Fasciola hepatica,* but the eyespot is considerably lighter than in that form. The time required for the development of the miracidium varies a little according to the season of the year. In the summer time it generally takes from two to three weeks.

In the summer of 1920 I was able to prove experimentally that two species of snails, viz., *Planorbis coenosus* Benson (Fig. 13) and *Segmentina largillierti* Dkr. (Fig. 14) could serve as the intermediate hosts of *Fasciolopsis buski.* When these species of snails were placed in water containing numerous actively swimming miracidia, the miracidia were soon to be seen swarming around the snails, trying to bore into the head, foot, tentacles, mantle and other parts. As the miracidia penetrated into the snails they threw off their coats of cilia. After penetration they became rounded and immobile, and losing the digestive tract are changed into mother sporocysts. The young mother sporocysts were elliptical in shape and measured 0.08 by 0.07 mm. (Fig. 4). These sporocysts gradually increase in size, the embryonic cells enter into cleavage and gradually rediae become recognizable within the sporocysts (Figs. 5 and 6). The rediae which develop within the sporocysts are cylindrical in shape. In the anterior region of the body, there is developed a ring shaped collar and near the posterior end are formed two lateral protuberances. The pharynx is large and the intestinal cavity is wide containing dark brown material. In the posterior part of the body many germ cells are visible. The redia at this stage of development (Fig. 7) escapes from the mother sporocysts and moves actively. These young rediae penetrate gradually into the walls of the alimentary canal, respiratory cavity and liver of the snail.

As time goes on the rediae increase in size and inside of them develop cercariae. In some of the rediae, however, a new generation of rediae develop. Full grown rediae (Fig. 8) measure from 1 to 1.5 mm. in length and about 0.23 mm. in width. The intestine is now smaller in proportion to the size of the rediae and its contents are deep black in color. The body wall of the redia generally becomes a light brown in color. The mature cercaria while still within the redia measures about 0.23 mm. in length and 0.13 mm. in breadth. It has a long tail which measures 0.4 to 0.5 mm. in length. The body of the cercaria is full of cytogenous glands which give it a dark appearance. At this period a few coarse granules appear near the lower ends of the two divisions of the excretory bladder. Each redia contains from 4 to 7 cercariae. As the cercariae become mature they escape from the redia and swim freely.

The mature cercariae (Fig. 9) have fewer pigment granules than the younger ones. They measure from 0.21 to 0.23 mm. in length and 0.12 to 0.15 mm. in breadth. The tail is long, measuring 2 or 3 times the body length. The cercaria is flattened and has the general shape of a tadpole. The oral sucker is subterminal and has a diameter of about 0.04 mm. The pharynx is spherical, immediately behind the oral sucker, and has a diameter of about 0.02 mm. The bifurcation of the intestinal ceca can be observed close behind the pharynx, but their course is difficult to make out. The divisions of the excretory bladder can be observed in a sinous course on each side of the body. They are very conspicuous on account of the presence of coarse, highly refractile granules, the largest of which have a diameter of 7μ. The undivided portion of the excretory bladder is very small, has an elliptical form which sometimes changes and is not always easy to make out. The ventral sucker is situated a little behind the middle of the body and has a diameter of about 0.03 mm. Soon after their escape from the snail the mature cercariae attach themselves to plants, where they become encysted.

The encysted cercaria (Fig. 10) has a diameter of 0.13 to 0.14 mm. The excretory bladder is filled with coarse granulus, peculiar to this species, which furnish a reliable differential character. The wall of the cyst consists of two layers, and has a thickness of 7μ. Around this cyst is attached some residue from the cercaria which is faintly yellowish. In the summer the cercariae mature in about 5 to 7 weeks after the infection of the intermediate host.

Stages of Development in the Final Host

I fed some dogs and pigs with the encysted cercariae of *Fasciolopsis buski* and studied their development. These experiments are still under way but I will record here the development as far as it has been worked out. Worms obtained from the small intestine of a young pig 20 days after feeding with the encysted stage measured after preservation in formalin 1 to 1.6 mm. in length and 0.48 to 0.58 mm. in width. They were flattened and leaf like in form and light colored. The oral sucker had a diameter of 0.15 mm. and the pharynx 0.12 mm. The ventral sucker was already located in front of the middle of the body and had a diameter of 0.3 mm. The reproductive organs were but slightly developed and could scarcely be distinguished. The surface of the body was thickly set with short spines embedded in the cuticula.

Worms 24 to 27 days after feeding to an experimental puppy measured when preserved in formalin 2 to 3 mm. in length and 1 to 2 mm. in breadth (Fig. 11). In the largest specimen the oral sucker was situated somewhat ventrally and has a diameter of 0.25 mm. Just behind this was the pharynx which was spherical with a diameter of 0.15 mm. The place of bifurcation of the intestinal ceca was just behind the pharynx, and they ran to the posterior end parallel to the sides of the body. For the posterior half of their length they took a zigzag course. The conspicuous, bell-like ventral sucker is now well forward and has a diameter of 0.6 mm. The reproductive organs were not yet well developed, but the position of the genital pore just in front of the ventral sucker could be made out. The tubular cirrus sac could be traced backward along the median line. The uterus was at this stage a straight tube, the posterior half of which was under the cirrus sac. The shell gland (Mehlis gland) was clearly visible at the posterior end of the uterus. The ovary could be seen as a small group of cells at the side of the shell gland. The testes, also, could be seen as faintly defined groups of cells one behind the other, behind the ovary. The vitellaria were not yet developed at this stage. The main stem of the excretory bladder could be seen to divide into two or three branches just behind the shell gland. The excretory pore was distinctly visible at the posterior tip. The surface of the body appeared to be covered with scales owing to the presence of short spines embedded in the cuticula. The parts of the central nervous system showed clearly on each side of the pharynx.

Worms obtained 25, 28 and 33 days after feeding the cysts to young pigs moved about rather actively in a leech like manner. Some of the more fully extended specimens measured as much as 10 mm. in length. The largest specimens were somewhat reddish in color. They varied in size measuring when preserved in formalin 2 to 5 mm. in length and 1 to 3 mm. in width. They agreed in structure with those obtained from the experimental dog, only some were a little further developed.

Worms obtained from the small intestines of pigs 60, 66 and 89 days after feeding, when preserved in formalin, measured 10, 12 and 18 mm. in length and 4, 5 and 8 mm. in width. The uteri of the larger ones contained many eggs and agreed morphologically with the adult described below. Even in the smaller ones the reproductive organs at this stage were well developed and although the vitellaria were apparently not yet completely developed, the uteri contained eggs.

Worms obtained from experimental pigs 90 and 93 days after the feeding of the encysted cercariae were already fully developed (Fig. 12) and were discharging eggs. When preserved in formalin they were somewhat contracted and measured 13 to 15 mm. in length and 6 to 8 mm. in width. Living specimens were red in color and moved actively with a leech-like movement. Fully extended living specimens measured 20 mm. in length by 10 mm. in width. The oral sucker was 0.4 mm. in diameter and the pharynx, 0.5 mm. The intestinal ceca have the same course as in the descriptions of the adults from man. The bell-shaped ventral sucker is large and conspicuous and measured 1 mm.

in diameter. The genital pore was visible near the anterior margin of the ventral sucker. The cirrus sac was median and long and cylindrical in shape. The ovary and and shell gland are at this stage situated near the middle of the body. The folds of the uterus partly fill the space between the ovary and the ventral sucker and obscure the posterior part of the cirrus sac. The ovary is dendritically branched like the antlers of a stag and is situated to the right of Mehlis' gland. The testes lie one behind the other and are very much branched. The vitellaria occupy the space along the sides of the body from the region of the ventral sucker to the posterior end. The eggs are light yellowish brown in color and measure 0.12 to 0.13 mm. in length and about 0.08 in width. These specimens, although somewhat below the average of the measurements of adult specimens of *Fasciolopsis buski* from man and the pig, agree in structure with the description of this species.

SUMMARY

The miracidia develop and escape from the eggs of *Fasciolopsis buski* in about two to three weeks in the summer time in Formosa. The cercariae are able to develop in two different species of snails, viz., *Planorbis coenosus* Benson and *Segmentina largillierti* Dkr. The most characteristic feature of the cercaria is the conspicuous limbs of the excretory bladder, situated along the sides of the body and filled with coarse granules. The encysted cercariae, which also have this same characteristic, are found attached to water plants. By feeding experimental animals (dogs and pigs) with the encysted cercariae, immature forms and sexually mature adults were obtained from the small intestines.

The life-cycle of *Fasciolopsis buski* closely resembles that of *Fasciola hepatica*. From the eggs which escape into the water, the miracidia hatch which invade the snail host. In the intermediate host the miricidia metamorphose into sporocysts and the development of rediae and cercariae follow. The mature cercariae escape into the water and become encysted on water plants, awaiting ingestion by their final host. The life cycle of *F. buski* as well as its mode of infection furnishes data for the prevention and control of the disease which this parasite produces in man.

In conclusion I wish to express my sincere thanks to Professor Miyajima for his kind assistance.

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EXPLANATION OF PLATE XXIV

Fig. 1.—Underdeveloped egg.

Fig. 2.—Egg containing fully developed miracidium.

Fig. 3.—Miracidium.

Figs. 4-6.—Stages in development of mother sporocyst.

Fig. 7.—Immature redia.

Fig. 8.—Fully developed rediae containing cercariae.

Fig. 9.—Mature cercaria, ventral view.

Fig. 10.—Encysted cercaria.

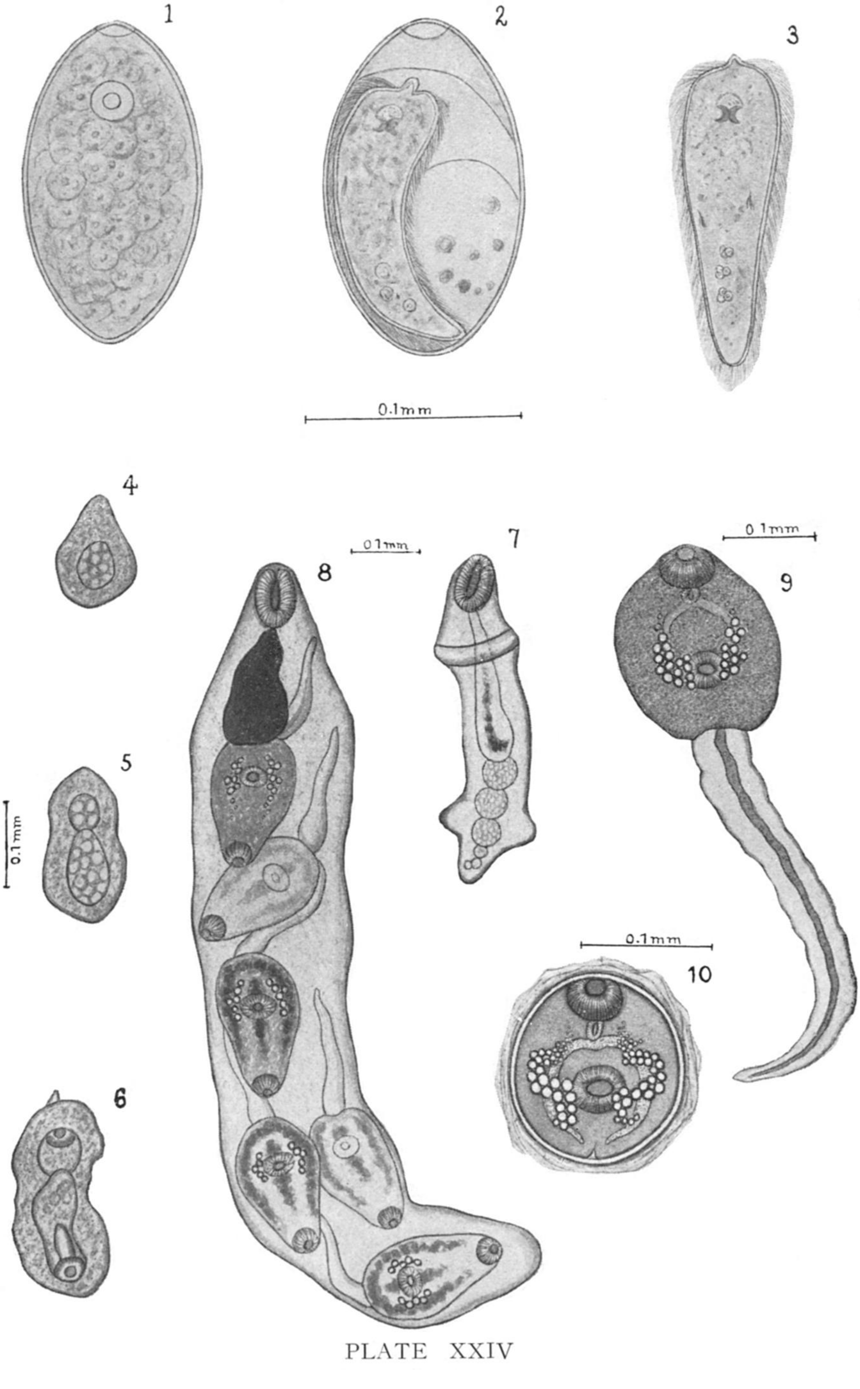

PLATE XXIV

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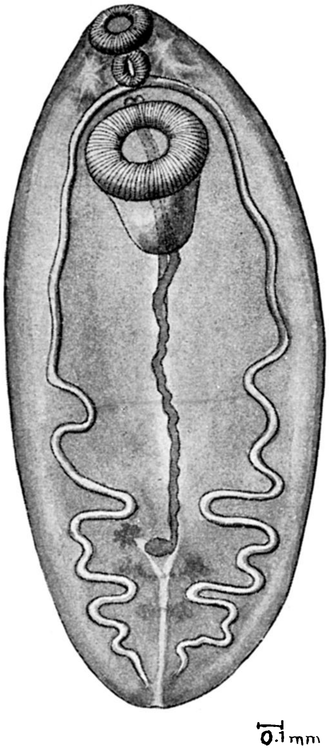

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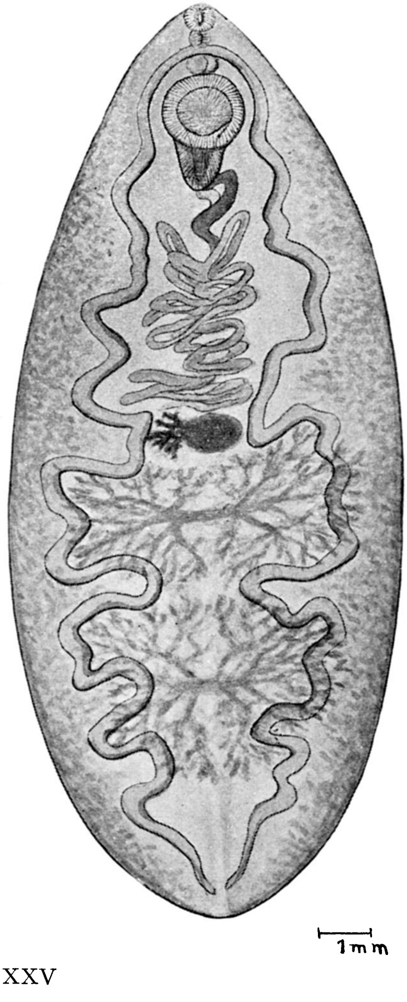

PLATE XXV

Fig. 11.—Immature worm, about 30 days after infection of final host.

Fig. 12.—Sexually mature worm, about 90 days after infection of final host.

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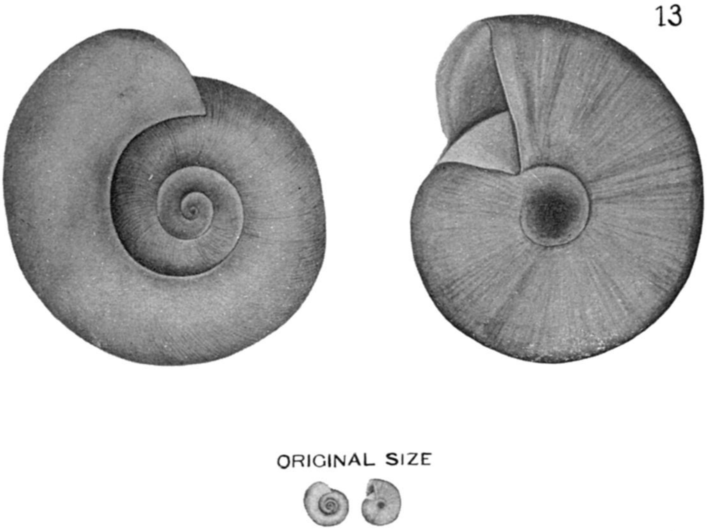

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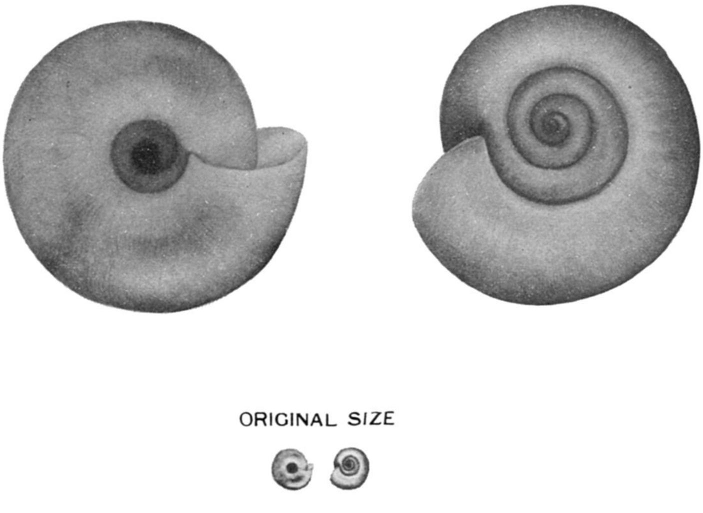

PLATE XXVI

Fig. 13.—*Planorbis caenosus* Benson, intermediate host of *F. buski.* ×5 and natural size.

Fig. 14.—*Segmentina largillierti* Dkr., intermediate host of *F. buski.* ×5 and natural size.